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Research Article
Growth, Yield and Physiological Characters of Three Types of Indonesian Rice Under Limited Water Supply

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Abstract

Background and Objective: Rice (*Oryza sativa* L.) is a staple food crop in Indonesia. Drought patterns are unpredictable and have complicated response mechanisms. This study aimed to determine the tiller number, stover yield, grain yield and physiological constituents (levels of antioxidants, proline, enzyme nitrate reductase (NR) and chlorophyll content) of rice that received limited supply of water. **Materials and Methods:** A 3×3 factorial design with three replications was used in this study. The first factor was the type of rice (Sidenuk, Way Apo or Pepe) and second factor was based on limited water field capacities of 75 and 100% of total field capacity and saturated water. Parameters observed were tiller number, stover yield, grain yield and physicochemical constituents (levels of antioxidants, proline, enzyme nitrate reductase (NR) and chlorophyll content). All data were analysed by using ANOVA, followed by Duncan multiple range test. **Results:** Lacking water conditions decreased the number of tillers by 39% for Sidenuk compared with that under saturated water condition. The number of tillers decreased by 48% for Way Apo and by 58% for Pepe. Limited water conditions decreased in yield and yield components of rice. The results showed that compared with saturated water, water limitations caused an increase in the antioxidant content of Sidenuk, Way Apo and Pepe seeds, with the increase reaching to 128, 80 and 61%, respectively. With less water, NR decreased by 89.6% in rice compared with that under saturated water condition. The total chlorophyll decreased with the increase in water limitation. Total chlorophyll of Sidenuk-LW decreased by 54.2% compared with that under Saturated Water (SW) and Sidenuk-FC decreased by 33.6% compared with that under Saturated Water (SW). The total chlorophyll of Way Apo-LW decreased 39.6% compared with that under the saturated water conditions. The total chlorophyll of Pepe-LW decreased 34.4% compared with that under the saturated water and Pepe-FC decreased 11.9% compared with that under the saturated water. **Conclusion:** The number of tillers, stover yield, grain yield and chlorophyll content of rice decreased with the increasing drought stress. The content of antioxidants, polyphenols and proline increased with the increasing drought stress.

Key words: Rice, water deficiency, antioxidants, polyphenols, proline

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Rice (*Oryza sativa* L.) is generally the source of carbohydrates and minerals for the majority of Indonesian people that accounts for 26.6% of total cereal acreage and 43.6% of the total food production[1]. The main challenges faced in the efforts to increase food production are (1) The increasing demand for rice production in accordance with increasing population, (2) Limited availability of global rice supplies and (3) Rising food prices. Rice is a staple energy source that is needed every day. The grain contains polyphenols in the form of phenolic acids, anthocyanins and proanthocyanidins, which have the nutraceutical ability and functional ingredients for health[2,3].

Lack of water supply is generally regarded as one of the limiting factors that affect plant productivity, physiological and biochemical processes in plants[4]. Drought causes cellular dehydration, which results in the release of water from the cytosol to the vacuole and apoplast. The response of plants to water stress involves changes in stomatal conductance, stunted growth, accumulation of osmolytes and the expression of specific genes[5]. Drought triggers a variety of plant responses, ranging from cell metabolism to changes in the rate of growth and yield. Understanding the biochemical and molecular responses to drought is important to know the mechanisms of plant resistance to water limited conditions[6]. Drought stress affects photosynthesis and assimilate translocation, where excessive dry stress results in plant death. Some plants respond to drought stress using various mechanisms of tolerance and avoiding stress[7,8].

With a limited supply of water for agriculture, the need for adapting rice to drought becomes important. Drought patterns are unpredictable and have complicated response mechanisms. It is difficult to determine the components needed to make plants resistant to drought. There is still limited information about the stress or dry rice paddies. This study aimed to determine the growth and physiological characteristics of rice under limited supply of water.

MATERIALS AND METHODS

Experimental location and materials: Experiment was conducted in the greenhouse of the Faculty of Agricultural and Animal Sciences at Diponegoro University from April-September 2016. The test soil was a 0-20 cm surface layer of oxisol. The measured soil traits indicated a clay texture with a soil pH of 6.75 and 0.12% N, 0.10% P_2O_5, 0.22% K_2O and 0.99% C. The varieties of rice used for the experiment were Sidenuk, Way Apo and Pepe. The growth season was approximately of 110 days. Approximately 15 kg amount of air-dried soil was placed in a plastic box with dimensions of $40\times30\times20$ cm^3.

Experimental design: The experiment was performed by using a 3×3 factorial design with three replications. The first factor was the three kinds of rice (Sidenuk, Way Apo and Pepe) and the second factor was the amount of water treatment field capacities of Lacking Water (LW), Field Capacity (FC) and Saturated Water (SW).

Measurement parameters and methods: The rice seeds were sown for 10 days in the soil medium and then transferred into a rectangular tub for a pot experiment (measuring $40\times30\times20$ cm^3). Each pot contained one seedling. Plants were fertilized with 300 kg ha^{-1} of N, 100 kg ha^{-1} of P_2O_5 and 100 kg ha^{-1} of K_2O. The field capacity was determined using gravimetric methods (42%). Plants were grown according the treatments.

Soil moisture levels were monitored for each treatment every week throughout the growing season by a gravimetric method. To maintain the expected level of dryness, appropriate amounts of water were added to each pot. The parameters measured were the number of tillers, stover yield, grain yield, weight of 100 grains, number of panicles, panicle filling, antioxidant activity[9], polyphenols[10], proline[11], Nitrate Reductase (NR) enzymes[12] and chlorophyll content[13].

Estimation of total antioxidant activity: The total antioxidant activity was evaluated using the 1,1-diphenyl-2-picryl-hydrazyl (DPPH) free radical scavenging and thiobarbituric acid reactive species (TBARS) methods[9]. The different concentrations samples, which had been previously prepared in 50% ethanol (100 L) were mixed with 50% ethanol (1.4 mL) and added to 0.004% DPPH (1 mL) in ethanol. The mixture was vigorously shaken and immediately incubated in the dark. After 70 min, the reaction reached a plateau. The absorbance of the reaction solution was determined using a UV-Vis spectrophotometer to monitor the absorbance at 517 nm. Ascorbic acid, a stable antioxidant was used as a positive control. The percentage of the DPPH radical-scavenging activity of the sample was calculated as follows:

$$\text{DPPH scavenging activity (\%)} = \frac{\text{A517 DPPH solution-A517 sample}}{\text{A517 DPPH solution}}\times 100$$

Determination of total phenolic compounds: Total phenolic compounds were determined according to Orak[10]. All samples and readings were prepared and measured in triplicate. Gallic acid was used as a standard. A stock standard solution of gallic acid (0.5 mg mL^{-1}) was prepared by dissolving 250 mg of dry gallic acid in 1 mL of extracting solvent and then diluted in 500 mL of distilled water. The stock solution was stored at 4°C. Working standards between 0.01 and 0.05 mg mL^{-1} were prepared by diluting the stock solution with distilled water. The extract was prepared with a concentration of 1 mg mL^{-1}. Approximately 100 µL of the extract was transferred into a test tube and 0.75 mL of Folin-Ciocalteu reagent (previously diluted 10-fold with deionized water) was mixed in it. The mixture was allowed to stand at room temperature for 5 min. Then, 0.75 mL of 6% (w/v) sodium carbonate was added to the mixture and mixed gently. After standing at room temperature for 90 min, the absorbance was read at 725 nm using a UV-Vis spectrophotometer. A standard calibration curve of gallic acid was plotted (0.01–0.05 mg mL^{-1}). The total phenolic contents were expressed as milligrams of gallic acid equivalents per gram (mg GAE g^{-1} extract). All measurements were obtained in triplicate.

Determination of proline content: Proline was measured according to the method of Bates[11]. Proline accumulation was determined by weighing 100 mg samples of leaf powder dry extract with 10 mL of sulfosalicylic acid solution (3%) centrifuged at 3500 rpm for 10 min. Two milliliters of the supernatant were pipette reacted with a 2 mL solution of ninhydrin acid (1.25 g of ninhydrin, 30 mL of glacial acetic acid, 20 mL of acid phosphate (6 M) in a terproline). Standard curve was constructed based on concentrations of 10-50 µg mL^{-1}. Trials were performed on three replications.

Measurement of nitrate reductase activity: Nitrate Reductase (NR) activity was measured by using a modified method of Krywult and Bielec[12]. Approximately 500 mg of the leaves of the plant shoots were sliced into small pieces and put in dark film canisters filled with 5 mL of phosphate buffer solution at pH 7.5. Both salts were heated at 110-130°C for 2 h. After 24 h of immersion, the buffer solution was replaced and 0.1 mL of KNO_3 was added as a substrate. Incubation after the addition of KNO_3 was applied for 2 h. Dye reagent comprising 0.2 mL of 1% sulfanilamide in 3N HCl and 0.2 mL of 0.02% N-naphthyl ethylenediamine were put in a test tube. In the test tube containing reagent dye, 0.1 mL of the filtrate from a dark tube was added and the reaction of NO_2 was allowed to proceed until the mixture turned into pink. After the color change, 2.5 mL of distilled water was added, the mixture was transferred into a spectrophotometer cuvette and the absorbance was observed at a wavelength of 540 nm.

Determination of photosynthetic pigment contents: The contents of total chlorophyll (chlorophyll a and b) were determined spectrophotometrically according to the Arnon method[13]. A fresh sample of leaves was placed in 85% (v/v) aqueous acetone. The extract was centrifuged at 4,000×g for 10 min. The supernatant was then removed and diluted with 85% aqueous acetone to a suitable concentration for spectrophotometric measurements. The absorbance was measured against a blank of pure 85% aqueous acetone at wavelengths of 644 and 663 nm. The amounts of photosynthetic pigments were expressed as mg g^{-1} fresh weight.

Data analysis: Data were analyzed using one-way analysis of variance followed by the Duncan Multiple Range Test[14] for comparisons among means with a significance level of 5%.

RESULTS AND DISCUSSION

Growth and yields of three types of rice under limited water supply: Statistical analysis of the data indicated that rice types, and limited water interaction had significant effects on number of tiller per plant, stover yield, grain weight and weight of 100 seeds. However, rice types and limited water had significant effects on the number of panicles per plant and panicle filled. Rice types had no significant effect on the number of panicles per plant and panicle filled. Limited water had significant effect on the number of panicles per plant and panicle filled (Table 1). Limited water conditions resulted in a decrease in the number of tillers by 39% for Sidenuk under saturated water conditions. The number of tillers decreased by 48% for Way Apo and by 58% for Pepe. The Way Apo seed, which got saturated water, showed the highest tiller number (46) compared to all other treatments. which got saturated water compared to all treatments applied. The Pepe tiller number (13) with the LW was the lowest number of tillers. The Way Apo tiller number differed from those of Sidenuk and Pepe. The tiller numbers are a characteristic of rice growth.

Sayar *et al.*[15] reported that water loss can reduce leaf water potential, which is followed by a decrease in turgor, stomata conduction and photosynthesis, thus reducing growth and crop yield. Ahmed *et al.*[16] reported that on cotton plant, the plant at height, root length, fresh and dry biomass, the number of leaves per plant and total leaf area were found to be decreased in drought stress compared with optimum irrigation plants.

Table 1: Number of tillers, results stover, grain weight, weight of 100 seeds, the number of panicles and panicle filled in response to limited water supply

Indonesian rice type	Drought stress	No. of tillers per plant	Stover yield (g)	Grain yield (g)	Weight of 100 grains (mg)	No. of panicles per plant	Panicle filled
Sidenuk	Lacking water	20.00^{f}	129^{e}	20.3^{d}	2.2^{f}	22.3^{c}	8.7^{cd}
	Field capacity	28.00^{d}	316^{c}	50.7^{c}	2.8^{d}	30.3^{bc}	9.0^{cd}
	Saturated water	33.00^{b}	478^{a}	190.7^{a}	4.2^{a}	53.0^{a}	39.0^{a}
Way Apo	Lacking water	24.00^{e}	87^{e}	60.7^{c}	1.8^{g}	19.6^{c}	3.7^{d}
	Field capacity	30.00^{c}	216.7^{d}	109.7^{b}	2.7^{d}	32.7^{bc}	19.3^{bc}
	Saturated water	46.00^{a}	390.7^{b}	184.7^{a}	3.6^{c}	44.3^{ab}	29.3^{ab}
Pepe	Lacking water	13.00^{g}	153.7^{e}	30.0^{d}	1.9^{g}	18.7^{c}	5.3^{d}
	Field capacity	24.00^{e}	241.7^{d}	119.0^{b}	2.5^{e}	55.0^{a}	22.3^{b}
	Saturated water	31.00^{bc}	457.3^{a}	182.0^{a}	3.9^{b}	45.0^{ab}	26.0^{b}
Sidenuk		27.22^{b}	307.6^{a}	87.2^{b}	3.03^{a}	35^{a}	18.9^{a}
Way Apo		33.77^{a}	231.4^{c}	118.3^{a}	2.69^{b}	32^{a}	17.4^{a}
Pepe		23.00^{c}	284.2^{b}	110.3^{a}	2.76^{b}	39^{a}	17.9^{a}
Lacking water		19.33^{c}	123.22^{c}	37.0^{c}	1.96^{c}	20^{b}	5.89^{c}
Field capacity		27.66^{b}	258.11^{b}	93.11^{b}	2.65^{b}	39^{a}	16.89^{b}
Saturated water		37.00^{a}	442.0^{a}	185.78^{a}	3.89^{a}	47^{a}	31.44^{a}
Rice types		166.67*	42.84*	34.04*	20.44*	1.48^{ns}	0.12^{ns}
Lacking water		441.42*	719.72*	736.31*	602.99*	21.21*	36.37*
Rice types × lacking water		18.59*	6.17*	22.89*	63*	3.10*	3.66*

$^{a-f}$Different letters in the same column show significant differences ($p<0.05$). *Values showing significant affect in the same column, ns: Non significant

Limited water conditions resulted in a decrease in yield and yield components of rice. The stover yield of the Sidenuk-LW decreased by 73% compared with Sidenuk-SW. The stover yield of Way Apo-LW decreased 78% compared with Way Apo-SW. The stover yield of Pepe-LW decreased 66% compared with Pepe-SW. The seed yield of Sidenuk-LW decreased by 89% compared with Sidenuk-SW, Way Apo-LW decreased by 67% compared with Way Apo-SW and Pepe-LW decreased 84% compared to Pepe-SW. The weight of 100 grains of Sidenuk-LW decreased by 48% compared with Sidenuk-SW, Way Apo-LW decreased by 50% compared with Way Apo-SW and decreased by 51% for Pepe-LW compared with Pepe-SW. Johnson and Henderson[17] examined the vegetative phase of growth after germination until just before flowering. In the phase where vegetative stems and leaves have formed, perfect assimilation activities and general changes in root reserves can occur as a result of assimilation.

The results showed that the number of panicle per plant of each variety was affected by limited water. The fewer the water supply less the amount of panicle per plant would be. The number of the panicle of Sidenuk-LW generally fell by 36.4% compared with that of Sidenuk-SW, the Way Apo-LW fell 55.8% compared with that of Way Apo-SW and Pepe-LW fell by 4.6% compared with that of Pepe-SW. Total panicle filled at Way Apo-LW were not significantly different with the panicles of Pepe-LW, Sidenuk-LW and Sidenuk-FC. Limited water conditions decreased the number of tillers by 39% for Sidenuk compared with saturated water conditions. The number of tillers of Way Apo-LW decreased by 48% compared with saturated water conditions and Pepe-LW decreased by 58% compared with saturated water conditions. Monkham *et al.*[18] documented the three selected genotypes for small yield reduction (less than 23%) had no delay in flowering, in fact, flowering was hastened, while those with large yield reduction (larger than 33%) had a flowering delay of at least 8 days. The yield reduction was 23-33% in the intermittent drought and 41-43% in the terminal drought. By comparing the mean values under different water availability conditions at each location, drought effect on tiller number production was negligible at both locations but terminal drought conditions reduced the mean panicle number and also reduced the individual panicle weight at both locations.

Plants that survive in drought condition has a mechanism with three main points: (a) The maintenance of a high plant water status during stress, (b) The maintenance of plant function at low plant water status and (c) The recovery of plant water status and plant function after stress[19]. The amount of tiller number do not necessarily generate much stover yield. The tiller number of Way Apo (46) with saturated water treatment produces less stover yield (390.7 g) than Sidenuk (478 g) and Pepe (457.3 g). Sidenuk and Pepe have the highest stover yield compared with other treatments.

Water supply shortage causes some reduction in the ability to form tillers, panicle number also caused reduced and incomplete filling seed or grain that was empty. Pepe rice panicle number at FC was not significantly different from the

Table 2: Level of antioxidant, polyphenols, proline, nitrate reductase, chlorophyll content of three rice types in response to limited water treatment

Rice types	Drought stress	Level of antioxidant (mg g^{-1})	Polyphenols (mg/100 g)	Proline (µmol g^{-1})	Nitrate reductase (µmol NO_2 g^{-1} per jam)	Chlorophyll content (mg g^{-1})
Sidenuk	Lacking water	12.53^{a}	42.7^{e}	7.17^{b}	1.03^{a}	1.36^{f}
	Field capacity	8.23^{c}	70.5^{bc}	3.84^{c}	4.10^{a}	1.97^{de}
	Saturated water	5.49^{d}	88.6^{a}	0.93^{de}	4.56^{a}	2.97^{a}
Way Apo	Lacking water	9.92^{b}	56.7^{d}	15.43^{a}	0.16^{a}	1.60^{ef}
	Field capacity	7.10^{c}	59.8^{cd}	3.84^{c}	2.53^{a}	1.99^{de}
	Saturated water	5.52^{d}	69.9^{bc}	0.65^{e}	6.16^{a}	2.65^{ab}
Pepe	Lacking water	11.39^{a}	49.4^{de}	4.64^{c}	0.16^{a}	1.60^{ef}
	Field capacity	8.18^{c}	61.0^{cd}	2.33^{d}	2.36^{a}	2.15^{cd}
	Saturated water	7.06^{d}	81.1^{ab}	1.4^{de}	2.73^{a}	2.44^{bc}
Sidenuk		8.75^{a}	67.3^{a}	3.98^{b}	3.23^{a}	2.10^{a}
Way Apo		7.51^{b}	62.1^{a}	6.64^{a}	2.95^{a}	2.00^{a}
Pepe		9.21^{a}	63.8^{a}	2.79^{c}	1.75^{a}	2.06^{a}
Lacking water		11.61^{a}	49.6^{c}	9.08^{a}	0.45^{b}	1.52^{c}
Field capacity		7.83^{b}	63.3^{b}	3.33.^{b}	3.15^{a}	2.03^{b}
Saturated water		6.02^{c}	79.9^{a}	0.99^{c}	4.33^{a}	2.68^{a}
Rice types		15.55*	1.3^{ns}	54.55*	0.81^{ns}	0.06^{ns}
lacking water		163.93*	45.46*	243.69*	5.21*	67.26*
Rice types × lacking water		5.56*	5.00*	49.51*	0.63^{ns}	3.25*

$^{a-f}$Different letters in the same column show significant differences (p<0.05). *Values showing significant affect in the same column, ns: Non significant

Sidenuk-SW, Way Apo-SW and Pepe-SW. It is probably that Pepe-FC on any conditions is capable of forming a panicle (55). However, when associated with a number of panicle fille, it only occupied 40.5%. The amount of panicle filled on Sidenuk-SW, Way Apo-SW and Pepe-SW was 73.6, 66.13 and 57.7%, respectively. Sidenuk has better grain quality for heaviest weight of 100 seeds (4.2 mg) with the results of the stover+highest seed (668.7 g), compared with other treatments (Table 1).

Drought stress induces a reduction in plant growth and the development of rice. Drought affects both elongation as well as expansion growth and it inhibits cell enlargement more than cell division[20]. Many aspects of plant growth are affected by drought stress, including leaf expansion, which is reduced due to the sensitivity of cell growth to water stress. Water stress also reduces leaf production and promotes senescence and abscission, resulting in decrease in the total leaf area per plant. A reduction in leaf area reduces crop growth and thus biomass production. Seed production is positively correlated with leaf area and it may also be reduced by leaf area reductions induced by drought stress[21].

In rice plants that are experiencing water stress, leaves roll up as a result of reduced leaf cell turgidity, thereby reducing the amount of CO_2 that diffuses into the leaf. This causes the transpiration rate to decrease, thereby reducing the supply of nutrients from the soil to the plant, the growth and the rice production compared with saturated water conditions. Nazar *et al.*[8] reported that cell division is sensitive to water shortage, which is related to a loss of turgor. Increased turgidity can stop cell division and result in smaller plants. The amount of chlorophyll is primarily related to the rate of photosynthesis. Chlorophyll is a photosynthetic apparatus that helps plants absorb energy from sunlight. When the photosynthesis rate is high, much water is used for photosynthesis before water vapor is released in the process. In addition, during grain filling, much water is used to move photosynthetic products from the source to the sinks, which are panicles (spikelets) filling with grain. The grain-filling rate (grain yield) depends on the source activity, sink capacity and the capability of carbohydrate accumulation[22].

Phytochemical compound and antioxidant content: Statistical analysis of the data indicated that rice types, lacking water and interaction had significant effects on the level of antioxidant and proline content. However, the interaction of rice types and limited water had significant effects on the polyphenols. Rice types had no significant effect on polyphenols. Limited water had significant effect on polyphenols. Interaction of rice types and limited water had no significant effects on NR. Rice types had no significant effect on NR but lacking water had significant effect on NR. Interaction of rice types and limited water had a significant effect on the chlorophyll content. Rice types had no significant effect on the chlorophyll content. Limited water had a significant effect on chlorophyll content (Table 2).

Results of this study showed that compared with saturated water conditions, water limitations caused an increase in the antioxidant content of Sidenuk, Way Apo and Pepe seeds, which increased by 128, 80 and 61%, respectively. The antioxidant content increased with increasing drought stress for all three varieties. Stress triggers the response of secondary metabolites and results in increased antioxidants. Amirjani and Mahdiyeh[6] noted that drought triggers a variety of plant responses, ranging from cell metabolism to changes in the rate of growth and yield. Understanding the biochemical and molecular responses to drought is important for understanding the mechanism of plant resistance to water-limited conditions.

The polyphenol content of grain also increased with increasing water stress by 52% for Sidenuk, 19% for the Way Apo and 39% for Pepe compared with saturated conditions. Drought stress caused an increase in proline content in the leaves of Sidenuk, Way Apo and Pepe, which increased by 671, 2274 and 231% compared with saturated conditions, respectively. Plants respond to drought stress through the status of water in the body by stabilizing the water potential, osmotic potential and turgor potential of cells. This condition affects the biosynthesis of osmotic compounds, such as proline and sugar. The accumulation of such compounds is reported to be an adaptive response to drought stress in different types of plants[23].

The results of this study showed that the higher level of drought stress (lacking water) resulted in higher accumulation of proline in the leaves of plants. Under stress conditions, plant cells have the ability to prevent decreases of water. Usually, the plants withstand such pressure by the accumulation of compatible solutions such as proline, which can help to bear the environmental stress. Drought affects the increase in proline in two ways: Increasing a number of enzymes used in proline synthesis and decreasing the activity of destructive enzymes of proline. A drop in turgor pressure is the first reason for the accumulation of proline due to drought stress. The amino acid proline is produced from the degradation of proteins in response to drought due to its compatibility with osmosis[24].

One of the mechanisms involved in acclimation seems to be an accumulation of proline, a compatible solute. Its concentration has been suggested as a general indicator of drought tolerance[25]. With less water, NR decreased by 89.6% in rice compared to saturated water conditions. The total chlorophyll decreased with increasing water limitations. Total chlorophyll of Sidenuk-LW decreased by 54.2% and Sidenuk-FC decreased by 33.6% compared to saturated water conditions. The total chlorophyll of Way Apo-LW decreased 39.6% compared with saturated water (SW) and Way Apo-FC decreased 24.9% compared with saturated water (SW). The total chlorophyll of Pepe-LW decreased 34.4% compared with saturated water (SW) and Pepe-FC decreased 11.9% compared with saturated water (SW). Drought can either directly or indirectly lead to growth retardation, death, accumulation of proteins and toxins, damaged biochemistry and enzyme inactivation. A reduction in water availability led to a significant reduction in leaf chlorophyll content[11]. Chlorophyll is very sensitive to drought-induced reduction in pigment content in many plant species. Drought causes the stomata to close, it decreases transpiration and chlorophylls content[26]. Drought stress decreases the amounts of both chlorophyll a and b. The decrease of total chlorophyll shows different effects in various cultivars[25]. Mild to severe drought can inhibit cell growth, cell wall synthesis, photo-chlorophyll formation, NR, ABA accumulation, cytokinins content, opening and closing of stomata, CO_2 assimilation, respiration and the accumulation of proline and sugars in plants[27]. Chlorophyll content of rice were 2.0-2.1 mg g^{-1} lower than chlorophyll content of *Brachiaria brizantha* Stapf (3.44 mg g^{-1}) that have nitrogen fertilizer[28] addition up to 150 kg N ha^{-1}.

CONCLUSION

It is concluded that less water conditions resulted in the decreasing number of tillers, stover yield and grain yield in rice. The total chlorophyll decreased with decreasing water supply. Moreover, the content of antioxidants, polyphenols and proline increased with decreasing water supply.

SIGNIFICANCE STATEMENTS

- Rice (*Oryza sativa* L.) is a staple food crop in Indonesia and also in the world
- Lack of water is generally regarded as one of the limiting factors that affect plant productivity, especially rice
- This study is useful in providing physiological basis that the antioxidants of rice under stress will add some important research results for rice as well as a functional food

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